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L9: Entry 1 of 3

File: PGPB

Oct 31, 2002

PGPUB-DOCUMENT-NUMBER: 20020160378

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20020160378 A1

TITLE: Stress-regulated genes of plants, transgenic plants containing same, and methods of use

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	Claims	KWIC	Draw Desc
Image												

☐ 2. Document ID: US 20020040490 A1

L9: Entry 2 of 3

File: PGPB

Apr 4, 2002

PGPUB-DOCUMENT-NUMBER: 20020040490

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20020040490 A1

TITLE: Expressed sequences of arabidopsis thaliana

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	Claims	KWIC	Draw Desc
Image												

☐ 3. Document ID: US 20020023280 A1

L9: Entry 3 of 3

File: PGPB

Feb 21, 2002

PGPUB-DOCUMENT-NUMBER: 20020023280

PGPUB-FILING-TYPE: new

DOCUMENT-IDENTIFIER: US 20020023280 A1

TITLE: Expressed sequences of arabidopsis thaliana

Full	Title	Citation	Front	Review	Classification	Date	Reference	Sequences	Attachments	Claims	KWIC	Draw Desc
Image												

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Terms	Documents
L8 and (barley or arabidopsis or oryza)	3

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'      (FILE 'HOME' ENTERED AT 11:15:51 ON 15 NOV 2002)

FILE 'REGISTRY' ENTERED AT 11:16:01 ON 15 NOV 2002
L1      1 S NICOTIANAMINE SYNTHASE/CN

FILE 'HCAPLUS' ENTERED AT 11:16:26 ON 15 NOV 2002

FILE 'REGISTRY' ENTERED AT 11:16:33 ON 15 NOV 2002
      SET SMARTSELECT ON
L2      SEL L1 1- CHEM :          2 TERMS
      SET SMARTSELECT OFF

      FILE 'HCAPLUS' ENTERED AT 11:16:33 ON 15 NOV 2002
L3      23 S L2
L4      19 S L3 (L) (BARLEY OR ARABIDOPSIS OR ORYZA)
L5      4 S L4 AND PD<19980430
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=> s 'nicotianamine synthase/cn

L1 1 NICOTIANAMINE SYNTHASE/CN

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=> d

L1 ANSWER 1 OF 1 REGISTRY COPYRIGHT 2002 ACS

RN 161515-44-2 REGISTRY

CN Synthase, nicotianamine (9CI) (CA INDEX NAME)

OTHER NAMES:

CN **Nicotianamine synthase**

MF Unspecified

CI MAN

SR CA

LC STN Files: AGRICOLA, BIOBUSINESS, BIOSIS, CA, CAPLUS, TOXCENTER

*** STRUCTURE DIAGRAM IS NOT AVAILABLE ***

19 REFERENCES IN FILE CA (1962 TO DATE)

19 REFERENCES IN FILE CAPLUS (1962 TO DATE)

=> d'ibib ab 1-4

L5 ANSWER 1 OF 4 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 1996:586037 HCAPLUS

DOCUMENT NUMBER: 125:239625

TITLE: A strategy for cloning the genes in the synthetic pathway of mugineic acid-family phytosiderophores

AUTHOR(S): Mori, S.

CORPORATE SOURCE: Faculty Agriculture, University Tokyo, Tokyo, 113, Japan

SOURCE: Genetic Manipulation of Crop Plants to Enhance Integrated Nutrient Management in Cropping Systems--1. Phosphorus, Proceedings of an FAO-ICRISAT Expert Consultancy Workshop, Patancheru, India, Mar. 15-18, 1994 (1995), Meeting Date 1994, 129-144. Editor(s): Johansen, C. International Crops Research Institute for the Semi-Arid Tropics: Patancheru, India.

CODEN: 63KAAB

DOCUMENT TYPE: Conference

LANGUAGE: English

AB Genes involved in the biosynthetic pathway of mugineic acid-family phytosiderophores were cloned. Initially, the genes for **nicotianamine synthase** and nicotianamine aminotransferase were confirmed to be induced by iron (Fe) deficiency and were partially purified. The partial amino acid sequences of the "d" peptide were detd. on 2D-PAGE, which appeared to be specific to Fe-deficient **barley** roots. Finally, seven Fe-deficiency specific clones were selected by "differential screening" of a cDNA library constructed from Fe-deficient **barley** roots and three DNA clones (Ids1, Ids2, and Ids3) were sequenced from amongst these. Strategies to clone the genes essential for the synthesis of phytosiderophores are discussed.

L5 ANSWER 2 OF 4 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 1996:202215 HCAPLUS

DOCUMENT NUMBER: 124:259698

TITLE: The role of nicotianamine synthase in response to Fe nutrition status in Gramineae

AUTHOR(S): Higuchi, Kyoko; Kanazawa, Kenji; Nishizawa, Naoko-Kishi; Mori, Satoshi

CORPORATE SOURCE: Dep. Appl. Biol. Chem., Univ. Tokyo, Tokyo, 113, Japan

SOURCE: Plant and Soil (1996), 178(2), 171-7

CODEN: PLSOA2; ISSN: 0032-079X

PUBLISHER: Kluwer

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Nicotianamine is an intermediate for the biosynthesis of mugineic acid-family phytosiderophores (MAS) in the Gramineae and a key substance for iron metab. in dicots. **Nicotianamine synthase** catalyzes the formation of nicotianamine from S-adenosylmethionine. **Nicotianamine synthase** activity was induced in **barley** roots at the 3rd day after withholding Fe supply and declined within one day following the supply of Fe³⁺-epihydroxymugineic acid. The induction **nicotianamine synthase** activity by Fe-deficiency was obsd. also in sorghum, maize, and rye, and the level of **nicotianamine synthase** activity was highly assocd. with the MAS secreted among graminaceous plant tested. Therefore, the **nicotianamine synthase** gene may be a suitable candidate for making a transgenic plant tolerant to Fe-deficiency.

L5 ANSWER 3 OF 4 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 1995:771685 HCAPLUS

DOCUMENT NUMBER: 123:165182

TITLE: Response of **nicotianamine synthase** activity to Fe-deficiency in tobacco plants as compared with **barley**

AUTHOR(S): Higuchi, Kiyoko; Nishizawa, Naoko-Kishi; Yamaguchi, Hirotaka; Roemheld, Volker; Marschner, Horst; Mori,

Satoshi
CORPORATE SOURCE: Fac. Agric. Agric. Life Sci., Univ. Tokyo, Tokyo, 113,
Japan
SOURCE: Journal of Experimental Botany (1995),
46(289), 1061-3
CODEN: JEBOA6; ISSN: 0022-0957
PUBLISHER: Oxford University Press
DOCUMENT TYPE: Journal
LANGUAGE: English

AB In vitro **nicotianamine synthase** activity was measured
in tobacco under Fe-deficient or Fe-sufficient conditions. Its activity
was not induced by Fe-deficiency, in contrast to **barley** roots,
implying that the mol. biol. regulation of **nicotianamine**
synthase in response to Fe-deficiency may be different between
tobacco and **barley**.

L5 ANSWER 4 OF 4 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 1995:319128 HCAPLUS
DOCUMENT NUMBER: 122:183289
TITLE: Purification and characterization of
nicotianamine synthase from
Fe-deficient **barley** roots

AUTHOR(S): Higuchi, Kyoko; Kanazawa, Kenji; Nishizawa,
Naoko-Kishi; Chino, Mitsuo; Mori, Satoshi
CORPORATE SOURCE: Lab. Plant nutrition Fertilizers, Univ. Tokyo, Tokyo,
113, Japan

SOURCE: Plant and Soil (1994), 165(2), 173-9
CODEN: PLSOA2; ISSN: 0032-079X

PUBLISHER: Kluwer
DOCUMENT TYPE: Journal
LANGUAGE: English

AB Nicotianamine (NA), the key precursor of the mugineic acid family
phytosiderophores (MAS), is synthesized from S-adenosylmethionine (SAM).
NA synthase was strongly induced by Fe-deficiency treatment, and the
activity increased to the max. level faster than the time of max. level of
MAS secretion and also before the appearance of severest chlorosis. The
enzyme was mainly localized in the roots of barley. NA synthase had an
optimum pH at 9.0, a mol. wt. of .apprx.40,000-50,000 estd. by gel
filtration or .apprx.30,000 by SDS-PAGE. Using hydrophobic chromatog.,
hydroxylapatite chromatog., and preparative SDS-PAGE, NA synthase was
purified as 1 band on SDS-PAGE.

=> d'14 ibib ab 1-19

L4 ANSWER 1 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 2002:482096 HCAPLUS

DOCUMENT NUMBER: 137:289578

TITLE: Nicotianamine synthase: Gene isolation, gene transfer and application for the manipulation of plant iron assimilation

AUTHOR(S): Douchkov, D.; Herbiak, A.; Koch, G.; Mock, H.-P.; Melzer, M.; Stephan, U. W.; Baeumlein, H.

CORPORATE SOURCE: Institut fuer Pflanzengenetik und Kulturpflanzenforschung (IPK), Gatersleben, D-06466, Germany

SOURCE: Plant and Soil (2002), 241(1), 115-119

CODEN: PLSOA2; ISSN: 0032-079X

PUBLISHER: Kluwer Academic Publishers

DOCUMENT TYPE: Journal; General Review

LANGUAGE: English

AB Basic cellular processes such as electron transport in photosynthesis and respiration require the precise control of iron homeostasis. To mobilize iron, plants have evolved at least two different strategies. The non-proteinogenic amino acid nicotianamine is an essential component of both pathways. We review the characterization of the **nicotianamine synthase** as a member of a novel class of enzymes, the cloning of the corresponding gene coding sequences of **barley**, **Arabidopsis** and tomato as well as the mol. basis of the chloronerva mutant exhibiting severe defects in the regulation of iron metab. Further, we report on current expts. utilizing various NAS-genes to manipulate iron assimilation in model and crop plants using transgenic sense and antisense approaches.

REFERENCE COUNT: 18 THERE ARE 18 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 2 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 2002:460905 HCAPLUS

TITLE: Identifying sugarcane expressed sequences associated with nutrient transporters and peptide metal chelators

AUTHOR(S): Figueira, Antonio; Kido, Ederson Akio; Almeida, Raul Santin

CORPORATE SOURCE: Centro de Energia Nuclear na Agricultura, Universidade de Sao Paulo, Piracicaba, 13400-970, Brazil

SOURCE: Genetics and Molecular Biology (2001), 24(1-4), 207-220

CODEN: GMBIFG; ISSN: 1415-4757

PUBLISHER: Sociedade Brasileira de Genetica

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Plant nutrient uptake is an active process, requiring energy to accumulate essential elements at higher levels in plant tissues than in the soil soln., while the presence of toxic metals or excess of nutrients requires mechanisms to modulate the accumulation of ions. Genes encoding ion transporters isolated from plants and yeast were used to identify sugarcane putative homologues in the sugarcane expressed sequence tag (SUCEST) database. Five cluster consensi with sequence homol. to plant high-affinity phosphate transporter genes were identified. One cluster consensus allowed the prediction of a full-length protein contg. 541 amino acids, with 81% amino acid identity to the Nicotiana tabacum NtPT1 gene, consisting of 12 membrane-spanning domains divided by a large hydrophilic charged region. Putative homologues to **Arabidopsis thaliana** micronutrient transporter genes were also detected in some of the SUCEST libraries. Iron uptake in grasses involves the release of the phytosiderophore mugeneic acid (MA) which chelate Fe³⁺ which is then absorbed by a specific transporter. Sugarcane expressed sequence tag (EST) homologous to genes coding for three enzymes of the mugeneic acid biosynthetic pathway [**nicotianamine synthase**; **nicotianamine transferase**; and putative mugeneic acid synthetase (ids3)] and a putative Fe³⁺-phytosiderophore transporter were detected. Seven sugarcane sequence clusters were identified with strong homol. to members of the ZIP gene family (ZIP1, ZIP3, ZIP4, IRT1 and ZNT1), while four

clusters homologous to ZIP2 and three to ZAT were found. Homologues to members of another gene family, Nramp, which code for broad-specificity transition metal transporters were also detected with constitutive expression. Partial transcripts homologous to genes encoding .gamma.-glutamylcysteine synthetase, glutathione synthetase, and phytochelatin synthase (responsible for biosynthesis of the metal chelator phytochelatin) and all four types of the major plant metal-chelator peptide metallothionein (MT) were identified: Type I MT being the most abundant (>1% of seed-library reads), followed by Type II which had a similar pattern of expression as that described for **Arabidopsis** MT. Identifying and understanding the expression of genes assocd. with nutrient uptake and metal tolerance could lead to the development of more nutrient-efficient sugarcane cultivars, or might allow the use of sugarcane as a hyper-accumulator plant for the restoration of contaminated areas in phytoremediation programs.

REFERENCE COUNT: 43 THERE ARE 43 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 3 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 2002:30851 HCAPLUS

DOCUMENT NUMBER: 136:178713

TITLE: Analysis of upstream region of **nicotianamine synthase** gene from **Arabidopsis**

AUTHOR(S): thaliana: presence of putative ERE-like sequence Suzuki, Kazuya; Nakanishi, Hiromi; Nishizawa, Naoko K.; Mori, Satoshi

CORPORATE SOURCE: Laboratory of Plant Molecular Physiology, Department of Applied Biological Chemistry, The University of Tokyo, Tokyo, 113-8657, Japan

SOURCE: Bioscience, Biotechnology, and Biochemistry (2001), 65(12), 2794-2797

CODEN: BBBIEJ; ISSN: 0916-8451

PUBLISHER: Japan Society for Bioscience, Biotechnology, and Agrochemistry

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Nicotianamine (NA) is present in all plants so far examd., and is thought to chelate transition metal ions. Previously, we isolated three **nicotianamine synthase** (NAS) genes of **Arabidopsis** thaliana (AtNAS1, 2, and 3) and showed that each NAS gene is differentially expressed. Deletion anal. of the 5' flanking region of AtNAS3 found a putative ethylene-responsive sequence, ATTTTCAAA.

REFERENCE COUNT: 20 THERE ARE 20 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 4 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 2001:840146 HCAPLUS

DOCUMENT NUMBER: 136:294231

TITLE: Increased iron efficiency in transgenic plants due to ectopic expression of nicotianamine synthase

AUTHOR(S): Douchkov, D.; Hell, R.; Stephan, U. W.; Baumlein, H.

CORPORATE SOURCE: Molecular Mineral Assimilation and Institut fur Pflanzengenetik und Kulturpflanzenforschung Gatersleben, Gatersleben, D-06466, Germany

SOURCE: Developments in Plant and Soil Sciences (2001), 92(Plant Nutrition), 54-55

CODEN: DVPSD8; ISSN: 0167-840X

PUBLISHER: Kluwer Academic Publishers

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The plant-endogenous chelator for some metal micronutrients including iron, nicotianamine (NA), plays an important role in symplasmic and phloem transport of iron by keeping it in soln. at cytoplasmic pH (Stephan and Scholz, 1996). Exogenous supply of NA for plants suffering from iron deficiency resulted in a re-greening by better iron distribution within the plants. The same was obsd. with the iron-accumulating tomato mutant chloronerva, the only plant known to be NA-free. Thus, the enzyme NA synthase (NAS) was isolated from roots of barley grown under iron deficiency. The corresponding gene was cloned and successfully expressed

in E. coli. Furthermore, NAS sequences were isolated from genomic DNA of tomato and A. thaliana. They were introduced by Agrobacterium in sense and antisense constructs in A. thaliana, tobacco and tomato. Tobacco sense lines overexpressing NAS from A. thaliana were selected based on significantly increased NA concns. in the leaves compared to wild type plants. Plants of these lines grew faster, were green instead of chlorotic and exhibited a higher iron efficiency than wild type plants under iron limiting conditions. Crop plants with NAS overexpression thus could provide higher and more stable yield on areas of iron deficiency.

REFERENCE COUNT: 10 THERE ARE 10 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 5 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 2001:580559 HCAPLUS

DOCUMENT NUMBER: 135:303367

TITLE: The expression of a **barley** HvNAS1
nicotianamine synthase gene

promoter-gus fusion gene in transgenic tobacco is induced by Fe-deficiency in roots

AUTHOR(S): Higuchi, Kyoko; Tani, Masaharu; Nakanishi, Hiromi; Yoshiwara, Toshihiro; Goto, Fumiyuki; Nishizawa, Naoko K.; Mori, Satoshi

CORPORATE SOURCE: Laboratory of Plant Molecular Physiology, Department of Applied Biological Chemistry, The University of Tokyo, Tokyo, 113-8657, Japan

SOURCE: Bioscience, Biotechnology, and Biochemistry (2001), 65(7), 1692-1696

CODEN: BBBIEJ; ISSN: 0916-8451

PUBLISHER: Japan Society for Bioscience, Biotechnology, and Agrochemistry

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Nicotianamine (NA) is a precursor for mugineic acid-family phytosiderophores, which are a crit. component of the Fe acquisition process in graminaceous plants. In addn., **nicotianamine synthase** (NAS) is strongly induced in these plants by Fe deficiency. NA is essential for Fe metab. also in dicots, but NAS is not induced by Fe deficiency. The authors introduced a **barley** HvNAS1 promoter-gus fusion gene into tobacco. GUS activity was induced in the roots of these plants by Fe deficiency, and was constitutively expressed at a low level in their leaves.

REFERENCE COUNT: 21 THERE ARE 21 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 6 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 2001:509590 HCAPLUS

DOCUMENT NUMBER: 136:197099

TITLE: Analysis of transgenic rice containing **barley**
nicotianamine synthase gene

AUTHOR(S): Higuchi, Kyoko; Takahashi, Michiko; Nakanishi, Hiromi; Kawasaki, Shinji; Nishizawa, Naoko K.; Mori, Satoshi

CORPORATE SOURCE: Laboratory of Plant Molecular Physiology, Department of Applied Biological Chemistry, The University of Tokyo, Tokyo, 113-8657, Japan

SOURCE: Soil Science and Plant Nutrition (Tokyo, Japan) (2001), 47(2), 315-322

CODEN: SSPNAW; ISSN: 0038-0768

PUBLISHER: Japanese Society of Soil Science and Plant Nutrition

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Nicotianamine (NA) is an intermediate in the biosynthetic pathway of mugineic acid-family phytosiderophores (MAs), which are essential components of the iron acquisition mechanism of graminaceous plants. NA is also considered to be an essential chelator of metal cations in dicots. Thus, NA and **nicotianamine synthase** (NAS, EC 2.5.1.43) play important roles in Fe metab. and homeostasis in higher plants. To evaluate the contribution of NAS to MAs biosynthesis, we introduced the **barley** hvnas1 gene under the control of the CaMV35S promoter into rice. Transgenic rice contg. a **barley** HvNAS1 genomic DNA

fragment was also analyzed. Although significant expression of hvnas1 driven by the CaMV35S promoter was confirmed by Northern and Western analyses, total NAS activity, endogenous NA, and 2'-deoxymugineic acid (DMA) secretion were not higher under Fe-deficient conditions compared to vector control plants. However, under Fe-sufficient conditions, the amts. of endogenous NA and secreted DMA increased slightly. No accumulation of NA due to an altered balance of nicotianamine aminotransferase (NAAT) and NAS activities was obsd. These results indicate that the NAS activity may be strictly regulated.

REFERENCE COUNT: 21 THERE ARE 21 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 7 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 2001:132604 HCAPLUS

DOCUMENT NUMBER: 135:353484

TITLE: **Nicotianamine synthase** gene
expression differs in **barley** and rice under
Fe-deficient conditions

AUTHOR(S): Higuchi, Kyoko; Watanabe, Shunsuke; Takahashi,
Michiko; Kawasaki, Shinji; Nakanishi, Hiromi;
Nishizawa, Naoko K.; Mori, Satoshi

CORPORATE SOURCE: Laboratory of Plant Molecular Physiology, Core
Research for Evolutional Science and Technology
(CREST), Japan Science and Technology Corporation,
Saitama, 332-0012, Japan

SOURCE: Plant Journal (2001), 25(2), 159-167
CODEN: PLJUED; ISSN: 0960-7412

PUBLISHER: Blackwell Science Ltd.

DOCUMENT TYPE: Journal

LANGUAGE: English

AB **Nicotianamine (NA)** is an intermediate in the biosynthetic pathway of the mugineic acid family phytosiderophores (MAs), which are crucial components of the iron acquisition app. of graminaceous plants. In non-graminaceous plants, NA is thought to be an essential chelator for metal cation homeostasis. Thus NA plays a key role in Fe metab. and homeostasis in all higher plants. **Nicotianamine synthase (NAS, EC 2.5.1.43)** catalyzes the trimerization of S-adenosylmethionine to form one mol. of NA. **Barley**, a plant that is resistant to Fe deficiency, secretes large amts. of MAs, whereas rice, a plant that is susceptible to Fe deficiency, secretes only small amts. In this study we isolated a genomic fragment contg. HvNAS1 from **barley** and three rice cDNA clones, osnas1, osnas2 and osnas3, from Fe-deficient rice roots. We also isolated a genomic fragment contg. both OsNAS1 and OsNAS2. In contrast to **barley**, in which Fe deficiency induces the expression of NAS genes only in roots, Fe deficiency in rice induced NAS gene expression in both roots and chlorotic leaves. The amts. of endogenous NA in both the roots and leaves were higher than in **barley**. We introduced **barley** genomic DNA fragments contg. HvNAS1 with either 9 or 2 kb of the 5'-flanking region into rice, using Agrobacterium-mediated transformation. Fe deficiency induced HvNAS1 expression in both roots and leaves of the transgenic rice, as occurs with rice NAS genes. **Barley** and rice NAS genes are compared in a discussion of alteration of the NAS genes during adaptation to Fe deficiency.

REFERENCE COUNT: 33 THERE ARE 33 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 8 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 2000:40345 HCAPLUS

DOCUMENT NUMBER: 133:54316

TITLE: Cloning of **nicotianamine synthase**
genes from **Arabidopsis thaliana**

AUTHOR(S): Suzuki, Kazuya; Higuchi, Kyoko; Nakanishi, Hiromi;
Nishizawa, Naoko K.; Mori, Satoshi

CORPORATE SOURCE: CREST, Japan Science and Technology Corporation (JST),
Tsukuba, 305-0047, Japan

SOURCE: Soil Science and Plant Nutrition (Tokyo) (1999),
45(4), 993-1002

CODEN: SSPNAW; ISSN: 0038-0768

PUBLISHER: Japanese Society of Soil Science and Plant Nutrition

DOCUMENT TYPE: Journal
LANGUAGE: English

AB **Nicotianamine synthase** (NAS) catalyzes the trimerization of S-adenosylmethionine to form one mol. of nicotianamine (NA). In order to identify the gene encoding NAS in dicotyledonous plants, **Arabidopsis thaliana** databases were searched using the nucleotide sequence of the NAS gene from **barley** (HvNAS), which was recently isolated. Several ESTs and 3 genomic sequences highly homologous to HvNAS were found in the databases. Based on these nucleotide sequences and that of HvNAS, 2 sets of primers were designed to isolate the NAS orthologs in **Arabidopsis** and 3 DNA clones encoding AtNAS (AtNAS1, 2, and 3) were obtained. These clones were expressed in *Escherichia coli* and their protein products displayed the NAS activity. The expression of AtNAS1 was detected in both shoots and roots of *A. thaliana* by RT-PCR; AtNAS3 expression was only detected in the shoots. In contrast, AtNAS2 expression was not detected in any organs.

REFERENCE COUNT: 40 THERE ARE 40 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 9 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 1999:753329 HCAPLUS
DOCUMENT NUMBER: 132:971
TITLE: Nicotianamine synthase genes, isolation, sequencing and use for agricultural and environmental purposes
INVENTOR(S): Baumlein, Helmut; Ganai, Martin; Herbig, Alexandra; Ling, Hong-qing; Mock, Hans-peter; Stephan, Udo
PATENT ASSIGNEE(S): Institut für Pflanzengenetik und Kulturpflanzen Forschung, Germany
SOURCE: PCT Int. Appl., 15 pp.
CODEN: PIXXD2
DOCUMENT TYPE: Patent
LANGUAGE: German
FAMILY ACC. NUM. COUNT: 1
PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9960107	A2	19991125	WO 1999-DE1585	19990518
WO 9960107	A3	20000323		

W: AE, AL, AM, AU, AZ, BA, BB, BG, BR, BY, CA, CN, CU, CZ, EE, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, RO, RU, SD, SG, SI, SK, SL, TJ, TM, TR, TT, UA, UG, US, UZ, VN, YU, ZA, ZW, AM, AZ, BY, KG, KZ, MD, RU, TJ, TM
RW: GH, GM, KE, LS, MW, SD, SL, SZ, UG, ZW, AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE, BF, BJ, CF, CG, CI, CM, GA, GN, GW, ML, MR, NE, SN, TD, TG

DE 19824307	A1	19991125	DE 1998-19824307	19980520
AU 9951507	A1	19991206	AU 1999-51507	19990518
BR 9902980	A	20010306	BR 1999-2980	19990719

PRIORITY APPLN. INFO.: DE 1998-19824307 A 19980520
WO 1999-DE1585 W 19990518

AB The invention relates to **nicotianamine synthase** genes, the isolation and use thereof. The invention can be used in agriculture and environmental protection. According to the invention, **nicotianamine synthases** and the genes coding for the latter are isolated and sequenced. Special embodiment examples are based on isolation from **barley** or tomatoes.

L4 ANSWER 10 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 1999:723152 HCAPLUS
DOCUMENT NUMBER: 131:348530
TITLE: Cloning of cDNA for plant nicotianamine synthase and use for preparation of transgenic plants tolerant to iron deficiency
INVENTOR(S): Mori, Satoshi; Higuchi, Kyoko; Suzuki, Kazuya; Nishizawa, Naoko; Nakanishi, Hiromi
PATENT ASSIGNEE(S): Japan Science and Technology Corp., Japan
SOURCE: PCT Int. Appl., 83 pp.

CODEN: PIXXD2
 DOCUMENT TYPE: Patent
 LANGUAGE: Japanese
 FAMILY ACC. NUM. COUNT: 1
 PATENT INFORMATION:

PATENT NO.	KIND	DATE	APPLICATION NO.	DATE
WO 9957249	A1	19991111	WO 1999-JP2305	19990430
W: AU, CA, JP, US				
RW: AT, BE, CH, CY, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE				
CA 2327261	AA	19991111	CA 1999-2327261	19990430
AU 9936273	A1	19991123	AU 1999-36273	19990430
EP 1077255	A1	20010221	EP 1999-918274	19990430
R: DE, ES, FR, GB, IT, NL				
PRIORITY APPLN. INFO.:		JP 1998-137685	A	19980430
		WO 1999-JP2305	W	19990430

AB **Nicotianamine synthase** (NAS1), an enzyme in the of mugineic acid biosynthesis pathway, was isolated and purified from *Hordeum vulgare* cultivated in a medium lacking iron. The cDNA encoding the enzyme was cloned by PCR using the primers derived from the partially detd. N-terminal amino acid sequence. Cloning of the cDNA for the enzyme from *Oryza sativa* and *Arabidopsis thaliana* was also described. Claimed are methods of recombinant prepn. of NAS1, (monoclonal) antibodies to NAS1, transgenic plants or fruits expressing NAS1, and methods of purifn. of NAS1 in the presence of thiol protease inhibitors such as E-64. This gene can be used for the prepn. of transgenic plants, e.g., Gramineae, highly tolerant to iron deficiency.

REFERENCE COUNT: 5 THERE ARE 5 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 11 OF 19 HCAPLUS COPYRIGHT 2002 ACS
 ACCESSION NUMBER: 1999:656714 HCAPLUS
 DOCUMENT NUMBER: 132:10326
 TITLE: Isolation, characterization and cDNA cloning of **nicotianamine synthase** from **barley**. A key enzyme for iron homeostasis in plants
 AUTHOR(S): Herbik, A.; Koch, G.; Mock, H.-P.; Dushkov, D.; Czihal, A.; Thielmann, J.; Stephan, U. W.; Baumlein, H.
 CORPORATE SOURCE: Institut fur Pflanzengenetik und Kulturpflanzenforschung (IPK), Gatersleben, D-06466, Germany
 SOURCE: European Journal of Biochemistry (1999), 265(1), 231-239
 CODEN: EJBCAI; ISSN: 0014-2956
 PUBLISHER: Blackwell Science Ltd.
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB Basic cellular processes such as electron transport in photosynthesis and respiration require the precise control of iron homeostasis. To mobilize iron, plants have evolved at least two different strategies. The nonproteinogenous amino acid nicotianamine which is synthesized from three mols. of S-adenosyl-L-methionine, is an essential component of both pathways. This compd. is missing in the tomato mutant chloronerva, which exhibits severe defects in the regulation of iron metab. We report the purifn. and partial characterization of the **nicotianamine synthase** from **barley** roots as well as the cloning of two corresponding gene sequences. The function of the gene sequence has been verified by overexpression in *Escherichia coli*. Further confirmation comes from redn. of the nicotianamine content and the exhibition of a chloronerva-like phenotype due to the expression of heterologous antisense constructs in transgenic tobacco plants. The native enzyme with an apparent Mr of .apprxeq. 105 000 probably represents a trimer of S-adenosyl-L-methionine-binding subunits. A comparison with the recently cloned chloronerva gene of tomato reveals striking sequence homol., providing support for the suggestion that the destruction of the

nicotianamine synthase encoding gene is the mol. basis
of the tomato mutation.

REFERENCE COUNT: 38 THERE ARE 38 CITED REFERENCES AVAILABLE FOR THIS
RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 12 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 1999:647368 HCAPLUS

DOCUMENT NUMBER: 132:76041

TITLE: Presence of nicotianamine synthase isozymes and their
homologues in the root of graminaceous plants

AUTHOR(S): Higuchi, Kyoko; Nakanishi, Hiromi; Suzuki, Kazuya;
Nishizawa, Naoko K.; Mori, Satoshi

CORPORATE SOURCE: Laboratory of Plant Molecular Physiology, Department
of Applied Biological Chemistry, The University of
Tokyo, Tokyo, 113-8657, Japan

SOURCE: Soil Science and Plant Nutrition (Tokyo) (1999),
45(3), 681-691

CODEN: SSPNAW; ISSN: 0038-0768

PUBLISHER: Japanese Society of Soil Science and Plant Nutrition

DOCUMENT TYPE: Journal

LANGUAGE: English

AB **Nicotianamine synthase** (NAS) catalyzes the synthesis
of nicotianamine, which is an intermediate in the biosynthetic pathway of
mugineic acid family phytosiderophores (MAS). Using polyclonal anti-NAS
antibodies and recombinant NAS proteins, five NAS isoenzymes and one NAS
homolog were identified in Fe-deficient **barley** roots using
two-dimensional electrophoresis followed by Western blot anal. Other
unidentified NAS homologues that were induced by Fe-deficiency were also
detected in **barley** roots. Western anal. enabled to detect NAS
homologues in wheat, oats, rice, maize, and sorghum roots. In
graminaceous species, both the amt. and no. of NAS homologues were
correlated with the total NAS activity and Fe-deficiency tolerance. The
NAS isoform patterns differed among the graminaceous plants.

REFERENCE COUNT: 15 THERE ARE 15 CITED REFERENCES AVAILABLE FOR THIS
RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 13 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 1999:461782 HCAPLUS

DOCUMENT NUMBER: 131:209894

TITLE: Map-based cloning of chloronerva, a gene involved in
iron uptake of higher plants encoding nicotianamine
synthase

AUTHOR(S): Ling, Hong-Qing; Koch, Gudrun; Baumlein, Helmut;
Ganal, Martin W.

CORPORATE SOURCE: Institute for Plant Genetics and Crop Plant Research,
Gatersleben, D-06466, Germany

SOURCE: Proceedings of the National Academy of Sciences of the
United States of America (1999), 96(12), 7098-7103

CODEN: PNASA6; ISSN: 0027-8424

PUBLISHER: National Academy of Sciences

DOCUMENT TYPE: Journal

LANGUAGE: English

AB The uptake of iron in plants is a highly regulated process that is induced
on iron starvation. In tomato, the mutant chloronerva exhibits
constitutive expression of iron uptake responses and intercostal
chlorosis. Biochem., chloronerva is an auxotroph for nicotianamine, a key
polyamine in plant iron uptake metab. The chloronerva gene has been
fine-mapped onto the long arm of chromosome 1 in a large segregating
tomato population and yeast artificial chromosome clones encompassing the
region were isolated by using flanking markers. A cosmid contig contg.
the chloronerva gene was established, and complementing cosmids were
identified by transformation into the mutant. The chloronerva transcript
was identified by cDNA isolation using the complementing cosmids. The
gene encodes a unique protein of 35 kDa. The mutant harbors a single base
change compared with the wild type. Based on enzyme activity and sequence
similarity to the coding DNA sequence of the purified **barley**
enzyme the chloronerva gene encodes the enzyme **nicotianamine**
synthase.

REFERENCE COUNT: 27 THERE ARE 27 CITED REFERENCES AVAILABLE FOR THIS

L4 ANSWER 14 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 1999:424322 HCAPLUS
 DOCUMENT NUMBER: 131:226058
 TITLE: Iron acquisition by plants
 AUTHOR(S): Mori, Satoshi
 CORPORATE SOURCE: Laboratory of Plant Molecular Physiology, Department of Applied Biological Chemistry, The University of Tokyo, Tokyo, 113-8657, Japan
 SOURCE: Current Opinion in Plant Biology (1999), 2(3), 250-253
 CODEN: COPBFZ; ISSN: 1369-5266
 PUBLISHER: Current Biology Publications
 DOCUMENT TYPE: Journal; General Review
 LANGUAGE: English

AB A review with 33 refs. In nongraminaceous plants, the FeII-transporter gene and ferric-chelate reductase gene have been cloned from **Arabidopsis thaliana**, whereas FeIII-reductase has not. In graminaceous monocots, the genes for mugineic acids (MAS) synthesis, nas (**nicotianamine synthase**) and naat (nicotianamine aminotransferase), have been cloned from **barley**, whereas the FeIII-MAS transporter gene is yet to be cloned. Transferrin absorption in *Dunaliella* has been reported, suggesting a phagocytotic (endocytotic) Fe-acquisition mechanism. Work to develop transgenic cultivars tolerant to Fe-deficiency in calcareous soils is now in progress.

REFERENCE COUNT: 33 THERE ARE 33 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 15 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 1999:150695 HCAPLUS
 DOCUMENT NUMBER: 131:1306
 TITLE: Cloning of nicotianamine synthase genes, novel genes involved in the biosynthesis of phytosiderophores
 AUTHOR(S): Higuchi, Kyoko; Suzuki, Kazuya; Nakanishi, Hiromi; Yamaguchi, Hirotaka; Nishizawa, Naoko-Kishi; Mori, Satoshi
 CORPORATE SOURCE: Laboratory of Plant Molecular Physiology, Department of Applied Biological Chemistry, The University of Tokyo, Tokyo, 113-8657, Japan
 SOURCE: Plant Physiology (1999), 119(2), 471-479
 CODEN: PLPHAY; ISSN: 0032-0889
 PUBLISHER: American Society of Plant Physiologists
 DOCUMENT TYPE: Journal
 LANGUAGE: English

AB **Nicotianamine synthase (NAS)**, the key enzyme in the biosynthetic pathway for the mugineic acid family of phytosiderophores, catalyzes the trimerization of S-adenosylmethionine to form one mol. of nicotianamine. The authors purified NAS protein and isolated the genes nas1, nas2, nas3, nas4, nas5-1, nas5-2, and nas6, which encode NAS and NAS-like proteins from Fe-deficient **barley** (*Hordeum vulgare* L. cv Ehimehadaka no. 1) roots. *Escherichia coli* expressing nas1 showed NAS activity, confirming that this gene encodes a functional NAS. Expression of nas genes as detd. by northern-blot anal. was induced by Fe deficiency and was root specific. The NAS genes form a multigene family in the **barley** and rice genomes.

REFERENCE COUNT: 36 THERE ARE 36 CITED REFERENCES AVAILABLE FOR THIS RECORD. ALL CITATIONS AVAILABLE IN THE RE FORMAT

L4 ANSWER 16 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 1996:586037 HCAPLUS
 DOCUMENT NUMBER: 125:239625
 TITLE: A strategy for cloning the genes in the synthetic pathway of mugineic acid-family phytosiderophores
 AUTHOR(S): Mori, S.
 CORPORATE SOURCE: Faculty Agriculture, University Tokyo, Tokyo, 113, Japan
 SOURCE: Genetic Manipulation of Crop Plants to Enhance Integrated Nutrient Management in Cropping Systems--1. Phosphorus, Proceedings ofan FAO-ICRISAT Expert

Consultancy Workshop, Patancheru, India, Mar. 15-18, 1994 (1995), Meeting Date 1994, 129-144. Editor(s): Johansen, C. International Crops Research Institute for the Semi-Arid Tropics: Patancheru, India. CODEN: 63KAAB

DOCUMENT TYPE:

Conference

LANGUAGE:

English

AB Genes involved in the biosynthetic pathway of mugineic acid-family phytosiderophores were cloned. Initially, the genes for **nicotianamine synthase** and nicotianamine aminotransferase were confirmed to be induced by iron (Fe) deficiency and were partially purified. The partial amino acid sequences of the "d" peptide were detd. on 2D-PAGE, which appeared to be specific to Fe-deficient **barley** roots. Finally, seven Fe-deficiency specific clones were selected by "differential screening" of a cDNA library constructed from Fe-deficient **barley** roots and three DNA clones (Ids1, Ids2, and Ids3) were sequenced from amongst these. Strategies to clone the genes essential for the synthesis of phytosiderophores are discussed.

L4 ANSWER 17 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 1996:202215 HCAPLUS

DOCUMENT NUMBER: 124:259698

TITLE: The role of nicotianamine synthase in response to Fe nutrition status in Gramineae

AUTHOR(S): Higuchi, Kyoko; Kanazawa, Kenji; Nishizawa, Naoko-Kishi; Mori, Satoshi

CORPORATE SOURCE: Dep. Appl. Biol. Chem., Univ. Tokyo, Tokyo, 113, Japan

SOURCE: Plant and Soil (1996), 178(2), 171-7

CODEN: PLSOA2; ISSN: 0032-079X

PUBLISHER: Kluwer

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Nicotianamine is an intermediate for the biosynthesis of mugineic acid-family phytosiderophores (MAS) in the Gramineae and a key substance for iron metab. in dicots. **Nicotianamine synthase** catalyzes the formation of nicotianamine from S-adenosylmethionine. **Nicotianamine synthase** activity was induced in **barley** roots at the 3rd day after withholding Fe supply and declined within one day following the supply of Fe³⁺-epihydroxymugineic acid. The induction **nicotianamine synthase** activity by Fe-deficiency was obsd. also in sorghum, maize, and rye, and the level of **nicotianamine synthase** activity was highly assocd. with the MAS secreted among graminaceous plant tested. Therefore, the **nicotianamine synthase** gene may be a suitable candidate for making a transgenic plant tolerant to Fe-deficiency.

L4 ANSWER 18 OF 19 HCAPLUS COPYRIGHT 2002 ACS

ACCESSION NUMBER: 1995:771685 HCAPLUS

DOCUMENT NUMBER: 123:165182

TITLE: Response of **nicotianamine synthase** activity to Fe-deficiency in tobacco plants as compared with **barley**

AUTHOR(S): Higuchi, Kiyoko; Nishizawa, Naoko-Kishi; Yamaguchi, Hirotaka; Roemheld, Volker; Marschner, Horst; Mori, Satoshi

CORPORATE SOURCE: Fac. Agric. Agric. Life Sci., Univ. Tokyo, Tokyo, 113, Japan

SOURCE: Journal of Experimental Botany (1995), 46(289), 1061-3

CODEN: JEOA6; ISSN: 0022-0957

PUBLISHER: Oxford University Press

DOCUMENT TYPE: Journal

LANGUAGE: English

AB In vitro **nicotianamine synthase** activity was measured in tobacco under Fe-deficient or Fe-sufficient conditions. Its activity was not induced by Fe-deficiency, in contrast to **barley** roots, implying that the mol. biol. regulation of **nicotianamine synthase** in response to Fe-deficiency may be different between tobacco and **barley**.

ACCESSION NUMBER: 1995:319128 HCAPLUS

DOCUMENT NUMBER: 122:183289

TITLE: Purification and characterization of
nicotianamine synthase from
Fe-deficient **barley** roots

AUTHOR(S): Higuchi, Kyoko; Kanazawa, Kenji; Nishizawa,
Naoko-Kishi; Chino, Mitsuo; Mori, Satoshi

CORPORATE SOURCE: Lab. Plant nutrition Fertilizers, Univ. Tokyo, Tokyo,
113, Japan

SOURCE: Plant and Soil (1994), 165(2), 173-9

CODEN: PLSOA2; ISSN: 0032-079X

PUBLISHER: Kluwer

DOCUMENT TYPE: Journal

LANGUAGE: English

AB Nicotianamine (NA), the key precursor of the mugineic acid family
phytosiderophores (MAs), is synthesized from S-adenosylmethionine (SAM).
NA synthase was strongly induced by Fe-deficiency treatment, and the
activity increased to the max. level faster than the time of max. level of
MAs secretion and also before the appearance of severest chlorosis. The
enzyme was mainly localized in the roots of barley. NA synthase had an
optimum pH at 9.0, a mol. wt. of .apprx.40,000-50,000 estd. by gel
filtration or .apprx.30,000 by SDS-PAGE. Using hydrophobic chromatog.,
hydroxylapatite chromatog., and preparative SDS-PAGE, NA synthase was
purified as 1 band on SDS-PAGE.